THE QUESTION OF THE STRUCTURAL RELATIONSHIP BETWEEN ANTIGEN AND ANTIBODY MOLECULES

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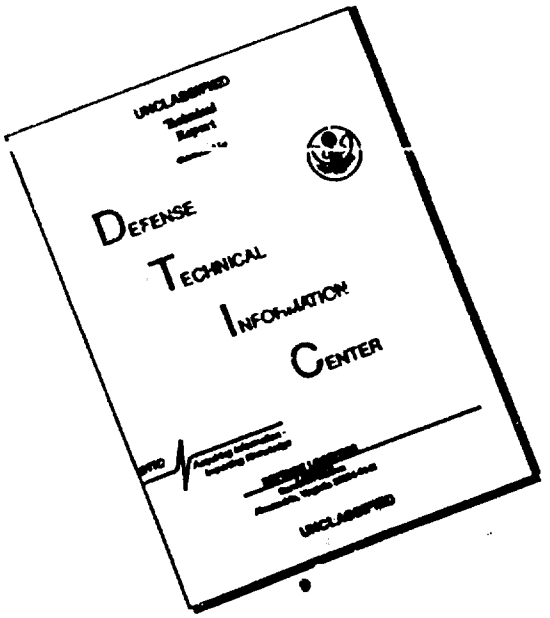

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O. Ye. Viazov and P. I. Çeitlin

(Presented by A. D. Speranski, Member of the Academy, March 12, 1956).

The mechanism of antibody production, and the reasons why antigens specifically unite with specific antibodies, form one of the central problems of present-day theoretical immunology.

Today, the leading position is occupied by the theory advanced in 1928 by N.F. Gamalei and subsequently developed by a number of research workers [1-5]. According to this theory, the molecules of the antibody are formed in the reticulo-endotheliar cells in response to the incidence in the cell of an antigen protein molecule, which serves, so to speak, as a pattern or die on which newly forming γ-globulin molecules are stamped out. The union of the antibodies thus formed with the homologous antigen takes place through an action of intermolecular attractive forces (of the van der Waals type), non-specific by nature. The specificity of this union is due to a specific spatial distribution, on the surface of the protein molecule, of active groupings so positioned that the molecules of homologous antigens on the one hand and antibodies on the other hand are, so to speak, mirror images of each other.

This theory, because of its reasonableness and logic, has achieved almost complete acceptance. But numerous attempts to provide it with a direct experimental demonstration have been unsuccessful, and to some extent the theory is still of hypothetical character.

We decided, in our work, to try to find some approach to the experimental solution of this problem. It seemed to us important, in the first place, to establish whether or not homologous antigens and antibodies do have a "mirror" relationship one to the other.

If the thesis of the structural "complementariness" of antigens and corresponding antibodies is correct, then one should expect that upon immunization of a rabbit the antibodies B (see Figure 1) will in their structural aspect be some kind of mirror image of the antigen A; when this antiserum is in turn used for immunization, the antibodies B now formed, as mirror images of their antigen, will structurally resemble the originally used antigen A.

Our experiment was conducted according to the scheme in Figure 2. As shown in this figure, a 2% solution of electrophoretically homogeneous human serum albumen, of mobility about $6.5 \cdot 10^{-5}$ cm^2 $sec^{-1}v^{-1}$ (veronal buffer, Ph 8.6, ion strength 0.1), was used as antigen for immunizing two rabbits.

The immunization was carried out according to the following schedule: 1 ml of the antigen was injected intravenously four times per week for a total of three such weekly cycles.* On the tenth day, blood was taken from a vein in the rabbits' ears and serum prepared from it. However, titration of these sera by the complement fixation reaction showed that the antibody titer was insufficiently high (1:320). Therefore the rabbits, thirteen days after the bleeding, were given three more 0.5 ml injections on alternate days. Titration of the sera then obtained showed an antibody titer of 1:1200. These sera were used as antigens for immunizing three chickens, as follows: Three 0.5 ml intramuscular injections on alternate days, and fourteen days later three more of the same injections (0.5 ml on alternate days). Seven days after the last injection, the fowls were totally bled. Titration of the sera, by the complement fixation reaction, showed an antibody content of titer 1:400.

The fowl sera thus obtained were then used to sensitize four guinea-pigs (0.1 ml subcutaneously). Four other guinea-pigs, as controls, were sensitized in the same way with normal sera from non-immunized fowl. On the 31st day after sensitization, the sensitized guinea-pigs were all given an 0.8 ml intravenous injection of a 2% solution of human serum albumen. The results of this experiment are shown in Table 1.

Table 1

Anaphylactic reaction in guinea-pigs sensitized with fowl sera, in response to intravenous injection of human albumen.

Guinea-pig No.	Sensitization			Final injection		
	Antigen and date of injection	Dose, ml	Reaction	Antigen and date of injection	Dose, ml	Reaction
1	Immune fowl serum,	0.1	-	2% solution of	0.8	+ +
2	July 30	0.1	-	human albumen,	0.8	+ +
3		0.1	-	August 31	0.8*	+
4		0.1	-		0.8	+
5	Normal fowl serum,	0.1	-	2% solution of	0.8	-
6	July 30	0.1	-	human albumen,	0.8	-
7		0.1	-	August 31	0.8	-

* Part of the antigen went under the skin.

* Beginning from the second cycle, the injections were made after desensitization of the rabbits by the Bezredka method.

As the results in Table 1 show, the guinea-pigs which were sensitized by serum from fowl immunized with the "anti-albumen" rabbit serum exhibited, in response to intravenous injection of human albumen, a distinct picture of anaphylactic shock (dyspnoea, sternutation, discharge of urine and faeces, intensified rubbing of nose with paws). At the same time, not one of the guinea-pigs sensitized with normal fowl serum showed any symptoms of anaphylactic shock in response to the intravenous injection of 2% human albumen solution.

The only explanation for these findings, it seems to us, is that the antibodies against human albumen formed in the immune rabbit possessed a stereochemical configuration of mirror-image character with respect to this antigen. The immunization of fowl with these antibodies led to the formation, in these fowl, of antibodies likewise of a mirror-image stereochemical configuration with respect to the rabbit-serum antibodies. Consequently the antibodies obtained from the fowl had a stereochemical configuration similar to the configuration of human albumen, as indeed was revealed by the anaphylactic reaction.

It might, however, be suggested that the sensitization of the guinea-pigs took place by the primary antigen's "getting through"; it might, so to speak, have been passaged through the rabbit and the chicken. To eliminate this suspicion, the following check was devised.

It is well known that a foreign protein introduced into an animal distributes itself very rapidly throughout the humoral channels and then gradually introduces itself into the organism.

According to the findings of K. I. Kotkov and T. V. Sayenko [6], when a foreign serum marked with I^{131} is injected into a rabbit, the radiation count per ml of rabbit serum one hour after the injection is less by 200-400 times than in the initial foreign serum. This is apparently due to ordinary physical dilution. Although the transfer of serum from rabbit to fowl and from fowl to guinea-pig was carried out 7-10 days after the last injection, we neglected the fact that in this time a considerable amount of the foreign protein (as much as 90-99%) would have been eliminated from the organism, and allowed only for the actual dilution of the antigen; in the rabbit a 200-fold dilution, and in the fowl, with its smaller weight taken into account, a 100-fold dilution. Simple calculation shows that under these circumstances 0.1 ml of the fowl serum may have a content of about 0.025 γ of the initial antigen.

Actually the antigen content will be many times smaller, since a considerable amount of it will be split during the time it is in the organism.

With the above as our starting point, we effected a control sensitization of two guinea-pigs with human albumen in the amount of 0.1 γ (exceeding by four times the figure theoretically found). Final inoculation of these rabbits with 2% human albumen solution gave distinctly negative results.

These findings do not by any means settle the whole intricate complex of questions connected with the structural interrelationships between antigen and specific antibody. Nevertheless we think that they may indicate routes of experimental approach to this problem.

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REFERENCES

1) F. Breinl, E. Haurowitz, Za. Physiol. Chem., 192, 45 (1930).

2) S. Mudd, Immunol., 23, 423 (1932).

3) J. R. Marrack, The Chemistry of Antigens and Antibodies, London, 1938.

4) J. Pauling, J. Am. Chem. Soc., 62, 2643 (1940).

5) L. Pauling, D. H. Campbell, D. Pressman, Proc. Nat. Acad. Sci., 27, 125 (1941).

6) K. I. KOTKOV, T. V. SAYENKO, Ukrayinski Biokhim. Zhurn., 27, 341 (1955).

A B C

Fig. 1. Suggested scheme for antibody formation. A - human albumen. B - antibody of anti-albumen serum. C - antibody of antiserum to anti-albumen serum.

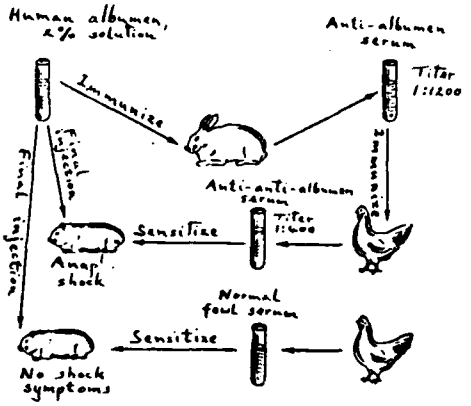

Fig. 2. Diagram of procedure in experiment on relationship between antigen and antibody molecules.